CONTINUOUS BLOOD GLUCOSE MONITORING AND DIETARY INTAKE IN A COHORT OF CHILDREN WITH SPINAL MUSCULAR ATROPHY TYPE II

by

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STATEMENT OF THESIS APPROVAL

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ABSTRACT

Previous studies have documented metabolic abnormalities in individuals with spinal muscular atrophy (SMA). This study investigated the impact of diet and fasting on blood glucose levels using a continuous blood glucose monitor (CGM) in conjunction with detailed food diaries in a small cohort (N=8) of children with SMA type II. Children ages 9 to 16 years, without any previous diabetes diagnoses or interventions, participated in the study. The study was completed in two parts; a one-time outpatient visit and a 4-day at-home portion. During the outpatient visit, fasting insulin, fasting glucose, and hemoglobin A1c (HgA1c) laboratory tests were obtained and the CGM was inserted. Anthropometric measurements including height, weight, triceps skin fold, and assessment of body composition using bioelectrical impedance analysis (BIA) were collected. Following the outpatient visit, participants tracked dietary intake and maintained the CGM for 4 days at home. For 1 day of the study, participants consumed a prescribed specialized diet comprised of 45.6% of total caloric intake from carbohydrates and adequate fiber intake (26.4g). Data analysis was both descriptive and exploratory. Seven of 8 participants were overfat according to BIA analysis. Although not statistically significant, blood glucose variation decreased 0.50 mg/dL for every gram of fiber consumed. Upon visual analysis, dietary intervention seemed to have a moderating effect on blood glucose levels of participants with varying metabolic control, as indicated by homeostatic model assessment for insulin resistance calculations. Additionally,

overfatness was common among participants and may correlate with blood glucose variability as BMI percentile increases. Implementing dietary recommendations for patients with SMA type II to increase daily fiber intake may prove beneficial for blood glucose regulation. Additionally, counseling to maintain adequate weight as assessed by both CDC BMI-for-age and body composition methods could improve patient blood glucose control in patients with SMA type II. Further research is required in order to determine significance of dietary intervention in children with SMA type II for the improvement of metabolic health.

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INTRODUCTION

Spinal muscular atrophy (SMA) is the most common genetic cause of death in infants with an incidence of 1 in 10,000 live births (1, 2). The autosomal recessive disease is caused by mutations or deletions within the survival motor neuron gene (*SMN1*) on chromosome 5q13. Two genes on chromosome 5q13 are responsible for SMN protein production: *SMN1*, the telomeric copy of the gene, and *SMN2*, the centromeric copy of the gene. Due to a mutation or deletion, *SMN1* can be rendered nonfunctioning. However, SMN protein production can be partially compensated for by the *SMN2* gene. The severity of SMA is dependent on the number of functioning copies of *SMN2* and the amount of SMN protein produced by these remaining genes. The disparity of SMN protein production leads to the degradation of α-motor neurons in the ventral horn of the spinal cord and brain stem nuclei, causing muscle atrophy and hypotonia. Variance in the amount and quality of SMN produced by *SMN2*, as well as other currently unknown modifying factors, allows for classification of SMA based on the highest motor milestone achieved (3).

SMA type I is the most commonly diagnosed form of SMA, comprising 50% of SMA diagnoses (1). Also known as Werdnig-Hoffman disease, SMA type I is the most severe form of SMA. Symptoms are present by 6 months of age and include inability to sit unaided and limited or inability to support the head. Other manifestations of SMA type I include bulbar dysfunction, swallowing, feeding, and gastrointestinal issues (4).

SMA type I has a survival rate of 18% past 4 years of age (5).

SMA type II symptoms usually become apparent between 7 and 18 months of age with expected survival into adulthood. Individuals with SMA type II are able to sit unsupported at some point during their clinical course but are never able to walk unaided (1, 6). Presentations of SMA type II may include bulbar weakness, issues chewing and swallowing, and commonly development of contractures and scoliosis (1). Likewise, frequent falls and exercise intolerance are noted effects of SMA type II (5). Additionally, individuals with SMA type II are at higher risk for respiratory infections due to insufficient clearance of mucus and secretions (4, 7).

Individuals diagnosed with SMA type III, also named Kugelberg-Welander disease, are able to stand and walk unaided for at least a period of their lives. SMA type III is diagnosed after 18 months of age with life expectancy into adulthood (8). Effects of SMA type III may include scoliosis and respiratory issues (4). Muscle and joint issues are also reported in this group.

Recently, there has been research suggesting a fourth sub-category of SMA, SMA type IV. SMA type IV is classified as adult onset SMA with diagnosis at 18 years of age or older, usually in the second or third decade of life. Individuals with SMA type IV have the ability to walk unaided at one point in their life (4, 8). Individuals with SMA type IV may also express minor motor impairment; however, they show few to no respiratory or gastrointestinal problems (4).

Aside from the manifestations of the disease, individuals with SMA experience a variety of associated symptoms that have detrimental nutritional and physical activity consequences. Specifically, these symptoms can affect weight, metabolism, and body

composition. Decreased lean muscle mass and disproportionately high fat mass may also play a role in the metabolic abnormalities seen in individuals with SMA. These abnormalities include metabolic acidosis, abnormal fatty acid metabolism, hyperlipidemia, hyperglycemia, and hypoglycemia when in a fasting state (9-11). Furthermore, these symptoms can be potentially traced to changes at the cellular level in individuals with SMA.

To explore the metabolic effects of SMA more thoroughly, Bowerman et al. examined whether metabolic and pancreatic defects would be seen in *SMN* depleted mice. The *SMN* depleted mice ($Smn^{+/-}$) did not present with phenotypic expression of SMA and produced approximately 50% of SMN protein in comparison to SMN production in wild type mice (10). The development of these mice may mimic less severe degrees of SMA in humans. $Smn^{+/-}$ mice at 1 year were 15% to 20% heavier than wild type mice which may indicate a role of the SMN protein in weight control and metabolism (10). Additionally, these mice presented with higher fasting glucose levels and hyperinsulinemia at 1 year (10).

In another study conducted by Bowerman et al., metabolic deviations were observed in SMN protein limited mice ($Smn^{2B/-}$), producing 15% of normal SMN protein quantities. Specifically, a temporal change in pancreatic composition from a normal ratio of insulin secreting β-cells to glucagon secreting α-cells to an abnormally low ratio of β-cell to α-cells in these mice was observed (12). Glucagon regulates gluconeogenesis; therefore, an abnormal increase of pancreatic α-cells may cause irregularities in gluconeogenesis and other metabolic processes. Similar pancreatic distributions were also found in infants with SMA type I at autopsy (12).

Additionally, β-cell function is important in the pathogenesis of type II diabetes mellitus. In a study by Butler et al., a statistically significant decrease in β-cell volume was observed at autopsy in obese adults with impaired fasting glucose or type II diabetes mellitus when compared to obese nondiabetic adults. Although different from the Bowerman et al. findings, where change in β-cell volume was potentially due to a fate switch from β-cell to α-cell characteristics, rather than increased apoptosis as found by Butler et al., both resulted in change of β-cell volume (13). Although different in pathology, the metabolic irregularities seen with changes in β-cell and α-cell volume further exemplify the effects of pancreatic alterations.

Muscle mass also has an important role in gluconeogenesis during fasting. Orngreen et al. demonstrated that during a 23-hour fasting period, 13 study participants with neuromuscular diseases, including 4 with SMA, had higher rates of lipolysis and contribution of glycerol from free fatty acids to gluconeogenesis than healthy controls. Additionally, all participants with SMA became hypoglycemic during the fasting period. This finding is likely the result of a lack of contribution of gluconeogenic substrates from muscle stores due to decreased lean body mass compared to healthy controls (11).

Additionally, individuals with SMA are at risk of developing increased fat mass and decreased fat free mass in comparison to healthy peers (14), often described as being overfat, or alternatively, metabolically obese. In a study by Sproule et al., fat mass index (kg fat mass/m^2) was found to be significantly increased in study participants with SMA, all types, compared to healthy children with consideration to age, race, and gender regardless of body mass index (BMI) (14). Additionally, a majority of the study population fell above the 85^{th} percentile on the fat mass index, classifying them as

overweight or obese in comparison to healthy peers (14).

Furthermore, Poruk et al. found increased fat mass in infants with SMA type I was associated with age (15). Also, infants with a weight-for-age greater than the 90th percentile had significantly higher percent fat mass than those within the 50th to 70th percentiles (15). The study also inferred that although children with SMA may fall within the underweight to healthy BMI ranges for the healthy pediatric population, children with SMA may have significantly higher fat mass than their peers (14, 15). Both of these studies demonstrate that individuals with SMA display an increase in fat mass and a decrease in fat free mass (14, 15). This trend was more extreme in participants with SMA type I and II and emphasizes the discrepancy between fat mass and BMI measurements in the SMA population (14).

Excess fat, or obesity, during childhood is the most prominent determinant for insulin resistance, metabolic syndrome, and type II diabetes (16-18). To further evaluate this mechanism along with other metabolic events in the SMA population, Hurst et al. conducted a pilot study exploring body composition, response to fasting, and glucose tolerance in children with SMA type II. Six children, ages 7 to 11 years, participated in the study. All 6 study participants were classified as obese as evaluated by the National Health and Nutrition Survey (NHANES) smoothed body fat percentiles for body composition (18). However, when charted in accordance to Centers for Disease Control and Prevention (CDC) BMI-for-age growth charts, only 3 of 6 participants were found to be obese, and 2 to be overweight, emphasizing the inadequacy of using CDC BMI-for-age percentiles as a measure of body composition for participants with SMA (18). Additionally, upon review of the food diaries submitted by the participants, participants

were not consuming excessive amounts of carbohydrates or calories. However, participants on average only met half of recommended dietary fiber intake needs.

The aforementioned study also explored glucose metabolism in children with SMA type II. Upon oral glucose tolerance testing, 3 of 6 participants met the American Diabetes Association's guidelines for impaired glucose tolerance (18). Five of 6 participants were insulin resistant as calculated by homeostatic model assessment for insulin resistance (HOMA-IR) during the supervised fast when compared to age- and gender-based guidelines (18). All participants exhibited hyperinsulinemia during fasting (18). Of note, hyperinsulinemia often anticipates glucose intolerance and may increase risk for type II diabetes in the future (19, 20). In summary, the pilot study indicated that SMA manifestations and progression may promote overfatness and affect glucose metabolism.

The culmination of negative metabolic symptoms, namely overfatness, abnormalities in glucose metabolism, and hyperinsulinemia may further exacerbate the effects of SMA and potentially lead to diseases such as type II diabetes and metabolic syndrome. Further exploration of the effects of the SMN protein on metabolic function is needed. Investigation of the outcomes of nutritional intervention in children with SMA type II has become of interest to increase quality of life, lifespan, and further deduce the roles of SMN protein in the human body.

The specific aims of this clinical study were to: 1) Monitor continuous blood glucose levels of 8 children with SMA type II 3 of whom have participated in a previous glucose tolerance and fasting study and have presented with hyperinsulinemia and; 2) Investigate and evaluate the impact of diet on continuous blood glucose levels of

participants under two conditions (typical diet and moderate carbohydrate/high-fiber). The hypothesis of this study was moderate carbohydrate intake in combination with increased fiber intake will better regulate blood glucose variation in comparison to regular diet.

METHODS

Participant Selection Criteria

The study enrolled 8 children diagnosed with SMA type II. The 6 children with SMA type II who previously participated in the pilot glucose study were recruited first. Due to limited response, additional children with SMA type II meeting inclusion criteria were recruited for a total of 8 participants enrolled.

Inclusion criteria included genetic diagnosis with SMA 5q, clinical diagnosis of SMA type II, and age 6-18 years. Exclusion criteria included acute illness, use of feeding tube for more than 50% of caloric intake, inability to swallow safely, medical diagnosis of type I or type II diabetes mellitus, and/or daily use of an oral hypoglycemic agent or insulin.

Experimental Plan

Children enrolled in the study were registered as outpatients in the University of Utah Center for Clinical and Translational Science (CCTS). During this visit, CGM devices were placed. The CGM devices were worn for 4 days at home. During these 4 days, the primary caregivers recorded the dietary intake of the children in a food diary. On 1 of the 4 days, the children abided by a prescribed specialized diet. On the fifth day, the CGM devices were removed by the primary caregivers and returned to the research group. The study protocol was approved by the Institutional Review Board (IRB 80144)

at the University of Utah.

Study Design

The study was completed in two parts: an outpatient visit overseen by trained personnel and an at-home portion completed over 4 days. For the outpatient visit, written informed parental consent and assent were obtained for all participants. Baseline measurements were collected including anthropometric measurements (weight, height, triceps skin fold, mid-arm circumference), bioelectrical impedance analysis (BIA), and standard vital signs. After an overnight fast, blood samples were obtained upon admission for hemoglobin A1c (HgA1c), blood glucose, and insulin. Following standard procedures, blood samples were analyzed by Associated Regional and University Pathologists, Inc. Laboratories (ARUP, Salt Lake City, Utah). The CGM devices (Medtronic iPro CGM, Northridge, California) were then placed on the children. Verbal explanation of operation of the CGM and how to take fingersticks using the provided glucometer (FreeStyle Lite Blood Glucose Monitoring System, 2010, Abbott Diabetes Care Inc., Alameda, California) was given. Following placement of the CGM and review of instructions, directions concerning diet and maintaining food diaries were provided by trained personnel. The children were instructed to consume their regular diet for 3 out of the 4 study days and on 1 day, abide by the prescribed specialized diet. A meal plan, cooking instructions, and all nonperishable items used in the prescribed specialized diet were provided in order to keep the diet as homogenous as possible amongst the participants.

Following the outpatient visit, children completed the remaining portion of the

study at home. Blood glucose values were automatically monitored by the CGM. All necessary blood glucose information was recorded by the device. Additionally, the participants recorded fingerstick glucometer values 3 to 4 times daily. These glucometer readings were used to ensure the CGM was properly calibrated. During the at-home portion, children followed their normal diet patterns for 3 of the 4 days and followed the prescribed specialized diet for 1 day. Primary caregivers completed food diaries documenting dietary intake for each day. After 4 days of tracking dietary intake and blood glucose monitoring, the CGM devices were removed and returned to the study office along with the completed food diaries.

Continuous Blood Glucose Monitoring

The CGM data were uploaded to the computer and calibrated using recorded blood glucose fingersticks. Compiled data were then analyzed for associations between the food diaries and carbohydrate intake. Variation was defined as the range of blood glucose values during the specified day.

Food Diary

The primary caregivers were responsible for recording participant dietary intake for 4 days through the use of a food diary. The food diary included types and quantities of foods, liquids, vitamins, and other supplements consumed. The food diaries were analyzed using Food Processor (version 10.5.2, 2009, ESHA Research, Salem, Oregon) for total kilocalories, macronutrient, and micronutrient content. The prescribed specialized diet day included 45.6% of calories from carbohydrate, 35.5% of calories

from fat, 18.9% of calories from protein, and contained 26.4 grams of fiber. Adherence to the prescribed specialized diet was evaluated and noted.

Anthropometric Measures

Anthropometric measurements were obtained by trained personnel during the outpatient visit using standard protocols (21). Weight was measured via a wheelchair accessible scale to the nearest tenth of a kilogram (Cardinal Scale Mfg. Co., Webb City, MO, kg). Height was evaluated through the use of segmental measures and arm span measures (standard, nonstretchable measuring tape, cm). Segmental measures included: head to shoulder, should to hip, hip to knee, knee to ankle, and ankle to heel. Ulnar measurements, chest circumference, and waist circumference were taken with a non-stretchable measuring tape (standard, nonstretchable measuring tape, cm). Triceps skin fold (Lange Skinfold Caliper, Cambridge, Maryland, mm) and mid-arm circumference (standard nonstretchable measuring tape, cm) were also assessed. All measurements were obtained from the right side of the body.

Weight status was determined using CDC BMI-for-age growth charts. Weight categorized between the 85th and 95th percentile was defined as overweight; weight categorized as the 95th percentile or above was defined as obese.

Bioelectrical Impedance Analysis

Body composition was measured using a bioelectrical impedance analysis machine (RJL Quantum Bioelectric Impedance Analyzer, Clinton Township, MI). Participants were reclined in the supine position on a hospital bed with electrodes

attached to their hands and feet. The current was sent through the body and the machine completed the process of measuring and calculating resistance. Recorded data were uploaded to the associated software (RJL Systems BC (Body Composition) Version 3.0.9, Clinton Township, Michigan). The software was then used to calculate fat mass percentage and fat free mass percentage per software automated pediatric equation. Body fat percentiles were assessed using smoothed body fat percentiles generated from data collected from NHANES dual energy x-ray absorptiometry (DXA) measurements of children ages 8-19 years (22).

HOMA-IR

Insulin resistance was determined using HOMA-IR calculations: [fasting glucose (mg/dL)(fasting insulin(μIU/mL))]/405 (23). Cut off values for insulin resistance were defined as ≥ 2.67 and ≥ 2.22 for prepubertal males and females, respectively; and ≥ 5.22 and ≥ 3.82 were used for pubertal males and females, respectively (23).

Statistical Analysis

Blood glucose levels taken by the CGM were compared to the recorded food diaries. Marked increases, decreases, relations, and patterns were noted. Blood glucose patterns observed during the prescribed specialized diet day were compared to the glucose patterns seen during the nonspecialized diet days as reported by the CGM.

The difference between the variations of participant blood glucose during the prescribed specialized diet day and the average variation in blood glucose over the 3 nonspecialized diet days was summarized and visually evaluated using diagramed data.

We noted the effects of the prescribed specialized diet day on blood glucose variation in comparison to the blood glucose variation during nonspecialized days. Additionally, the blood glucose response to food intake was evaluated by comparison of the food diaries and the CGM data.

Association between amount of fiber (in grams) consumed per day and variation in blood glucose per nonspecialized diet days was assessed using a linear mixed model. Pearson correlation tests were used to evaluate the associations between the following variables: 1) BMI percentile and variation in blood glucose on regular diet days; 2) Calories from carbohydrate and HOMA-IR; 3) Average percent of daily fiber recommendation intake during nonspecialized diet days and HOMA-IR; 4) Fat mass percentage and variation in blood glucose; 5) Fat mass percentage and HOMA-IR; 6) Correlation between fat free mass percentage and HOMA-IR. All analyses were performed with Statistical Analysis Software (version 9.4, SAS Institute Inc., 2013, Cary, North Carolina) and p-values <0.05 were considered statistically significant.

RESULTS

Eight children participated in the study; 3 females and 5 males. The mean age of participants was 12.1 ± 2.4 years (range 9-16 years). Within the participant population, 1 self-identified as black and 7 self-identified as Caucasian. Additionally, 3 children identified as Hispanic, 4 identified as non-Hispanic, and 1 participant did not specify ethnicity. All 8 children participated in the hospital portion of the study, completed the at-home segment, and returned the CGM to the study office along with completed food diaries.

Continuous Blood Glucose Monitoring

Within the group, per every 1 gram of fiber intake, blood glucose variation decreased by 0.50 mg/dL; however, this result did not reach statistical significance (p=0.22). Additionally, with every 1% increase of calories from carbohydrate, blood glucose variation increased by 0.073 mg/dL. This result did not reach statistical significance (p=0.76).

BMI percentile and variation in blood glucose on nonspecialized diet days had a moderate correlation; however, the association was not statistically significant (r=0.55, p=0.15). Within group, the correlation between fat mass percentage and variation in blood glucose was not strongly correlated (r= 0.20, p=0.67). An outlier with extremely

low body fat in comparison to the study population was excluded from this analysis.

Figure 1 depicts the difference in blood glucose variation observed from the average of nonspecialized diet days to the prescribed specialized diet day for all participants. Participant H was excluded due to digression from protocol. Additionally, Figures 2 and 3 depict differences in blood glucose variation for participants B and C on a day-by-day basis.

Food Diary

All 8 participants completed food diaries, with documentation of 3 days of regular diet patterns and 1 day of the prescribed specialized diet day. As a group, on average, the nonspecialized diet days were comprised of 50.5 ± 23.4% calories from carbohydrate, 33.3% ± 12.0% calories from fat, and 16.6% ± 5.3% calories from protein. Additionally, average grams consumed during the nonspecialized diet days were 195.5 ± 90.4g carbohydrate (range: 106.7g-393.0g), 56.8g ± 20.6g (range: 29.7g-97.3g) fat, and 64.3g ± 20.6g (range: 40.7g-91.7) protein. During the nonspecialized diet days, the percent of recommended grams of fiber consumption met was 52.1% ± 17.5% (range: 28.7%-64.4%) and the average of grams of fiber was 12.6g ± 4.7g (range: 7.1g-21.3g). Table 1 reports the group average calorie and macronutrient intake and percent of caloric intake during the nonspecialized diet days in comparison to the prescribed specialized diet.

Anthropometric Measurements and Bioelectrical Impedance Analysis

BMI, BMI classification, and BIA results are stated in Table 2. According to the CDC BMI-for-age growth charts, 5 of 8 participants were classified as overweight or

obese. Average fat mass percentage was 53.0% ± 8.9% (range: 32.3%-62.6%). Average fat free mass percentage was 47.1% ± 8.9% (range: 37.4%-67.7%).

HOMA-IR

HOMA-IR calculation results are stated in Table 2. Two of 8 participants were found to be insulin resistant based on HOMA-IR calculations. No significant correlation was found between the percent of calories from carbohydrate and HOMA-IR nor between the average grams of fiber intake during nonspecialized diet days and HOMA-IR ($r=0.15$, $p= 0.72$ and $r=0.19$, $p=0.65$, respectively). Significant associations were not found between fat mass percentage and HOMA-IR ($r=0.14$, $p=0.75$,) nor between fat free mass percentage and HOMA-IR ($r=0.14$, $p=0.74$).

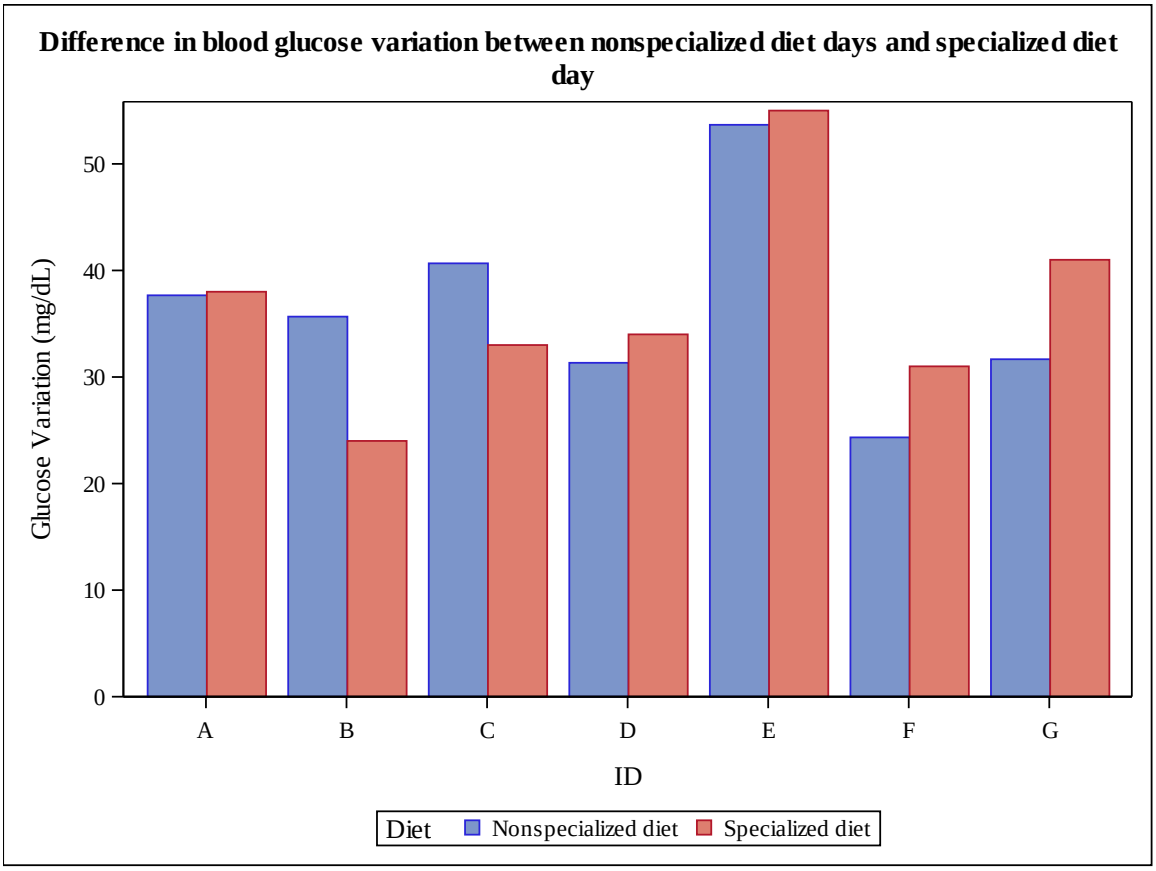


Figure 1. Data represent the difference in variation of blood glucose from average of nonspecialized diet days to prescribed specialized diet day.

* Participant H was excluded due to noncompliance with prescribed specialized diet protocol.

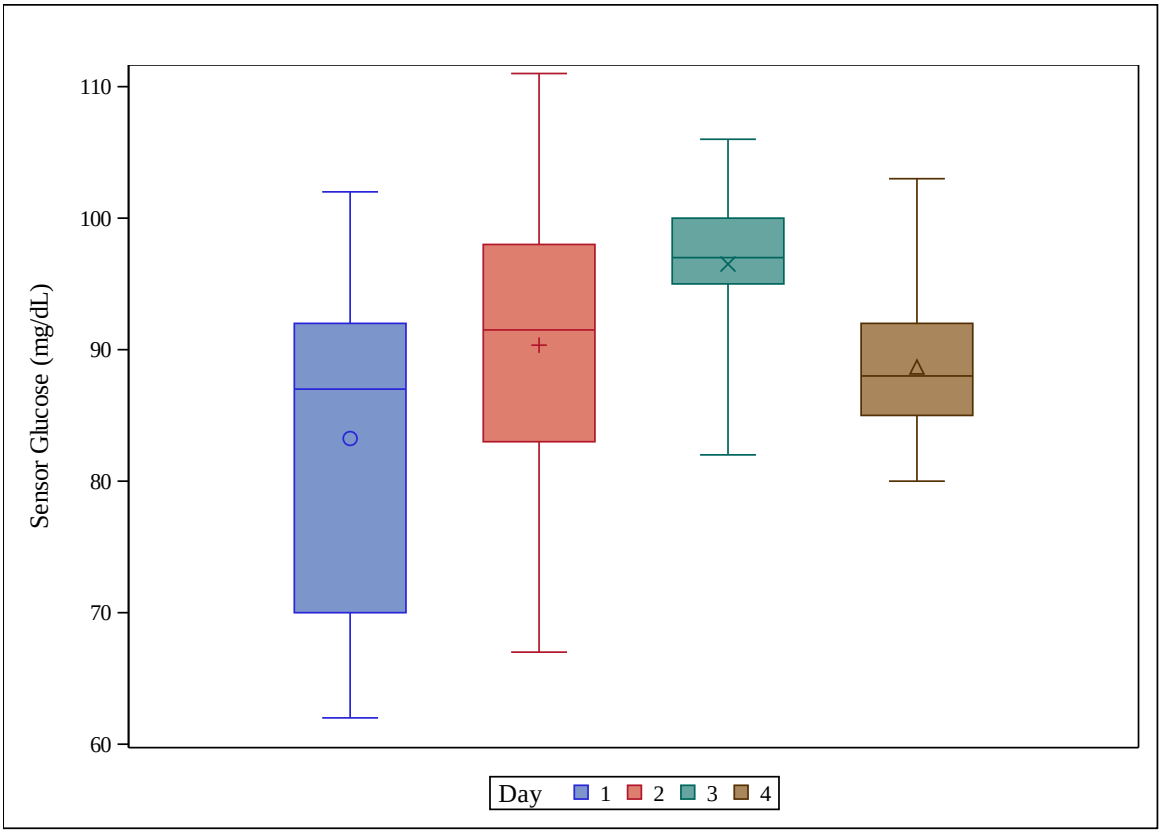


Figure 2. Data represent the change in variation of blood glucose day-by-day for participant B. Day 3 is the prescribed specialized diet day.

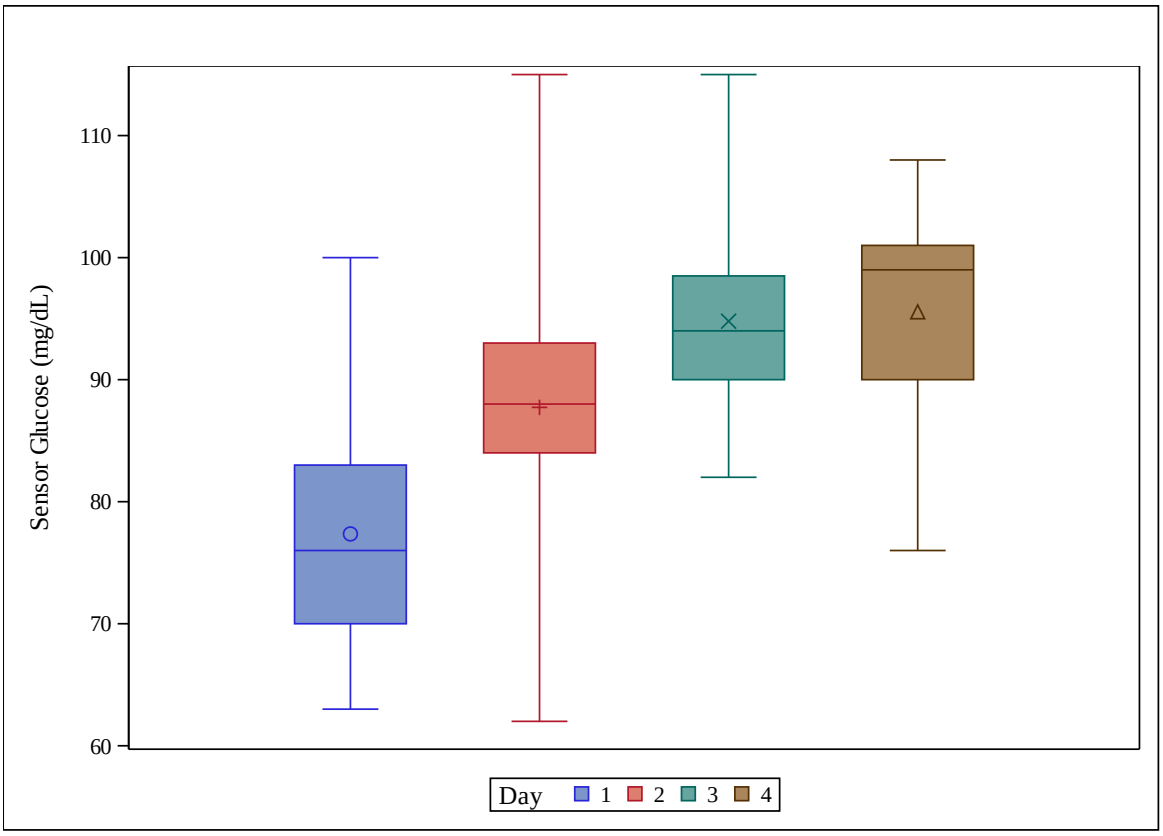


Figure 3. Data represent the change in variation of blood glucose day-by-day for participant C. Day 3 is the prescribed specialized diet day.

Table 1. Comparison of group average calorie and macronutrient intake and percent of caloric intake during the nonspecialized diet days to the prescribed specialized diet.

	Group Average of Nonspecialized Days	**Group Average Percent of Caloric Intake**	**Specialized Day**	**Percent of Caloric Intake**
Calories (kcal)	1548 ± 589.8	-	1196	-
Carbohydrate (g)	195.5 ± 90.4	50.5% ± 23.4%	138	45.6%
Fat (g)	56.8 ± 20.6	33.0% ± 12.0%	48	35.5%
Protein (g)	64.3 ± 20.6	16.6% ± 5.3%	57	18.8%
Fiber (g)	12.6 ± 4.7	3.3% ± 1.2%	26	8.8%

Table 2. Body composition data and HOMA-IR calculation results.

*Bold denotes out of normal reference range

† HOMA-IR cut off values are determined for obese children (>95%ile for BMI-for-age) (23)

‡ Denotes pre-pubertal status

Participant	Fat Mass %	Fat Free Mass %	BMI (kg/m^2)	BMI %ile	BMI z-score	HOMA-IR
A	53.7	46.3	23.8	**88**	1.15	3.12
B	62.6	37.4	22.6	66	0.41	1.7
C	55.6	44.4	26.7	**99**	**2.24**	**10‡**
D	52.3	47.7	20.7	**94**	1.52	**6.5‡**
E	32.3	67.7	11.1	**0**	**-7.4**	2.5
F	55.7	44.3	23.7	**93**	1.49	3.6
G	53.6	46.5	17.7	24	-0.71	3.3
H	57.8	42.2	24	**96**	1.77	4.5

HOMA-IR = [fasting glucose (fasting insulin)] /405		Male†	Female†
	Pre-pubertal	2.67	2.22
	Pubertal	5.22	3.82

DISCUSSION

As a prospective exploratory study, we were able to investigate the metabolic response of children with SMA type II to varying glucose loads and fiber intake over a short period of time. Unique findings include the decrease in blood glucose variation seen with increased fiber intake, the potential of dietary intervention having influence in children with a wide range of metabolic control, impaired blood glucose control with increased BMI percentile classification, and the consistency of overfatness in children with SMA type II despite normal BMI-for-age percentiles.

Although not statistically significant, the results indicate that an increased fiber load decreases variation in blood glucose in the participants studied. As reported, per every 1 gram of fiber intake, blood glucose variation decreased by 0.50 mg/dL. On the prescribed specialized diet day, participants, if protocol was followed, consumed 26 grams of fiber, meeting the RDA for the majority of our participant population. This amount contrasted vastly from observed intake during the nonspecialized diet days, ranging from 3 grams to 26 grams per day. On average, the participants met only 52% of recommended fiber intake, analogous to the findings of Hurst et al. in which participants were only consuming approximately one-half of recommended dietary fiber as reported by food diaries. To date, limited studies have been conducted on the effects of fiber intake on body weight in the pediatric population, with minimal research specific to the SMA community. One study, conducted by Brauchla et al., studied fiber intake in

relation to risk for overweight or obese status in the pediatric population. Study results indicated there was a significant decrease in risk of overweight/obesity status with increasing fiber intake in children ages 12 to 18, a similar age range as the participant population in this study (24). Moreover, for the current study, fiber intake during the prescribed specialized diet day was even higher than the highest tertile as reported by Brauchla et al., leaving potential for an even greater impact in individuals with SMA, especially if long term.

Another outcome of interest is the visual result that dietary intervention seems to have an impact on participants with varying metabolic control, as evaluated by HOMA-IR scores. Depicted in Figure 1, participants B and C had a notable decrease in blood glucose variation from the average of nonspecialized diet days in comparison to the prescribed specialized diet day. For participant C, lab values were indicative of insulin resistance as assessed by HOMA-IR, whereas lab values of participant B were not. As exemplified by the benefits shown in blood glucose control by both participants B and C, despite differing classification of insulin resistance, children with a wide spectrum of metabolic control and diagnosis of SMA may benefit from dietary intervention.

Additionally, although not statistically significant, results indicate blood glucose variation had a moderate correlation with BMI percentile on nonspecialized diet days. Therefore, study participants with increased BMI percentiles potentially had greater variation of blood glucose levels, indicating reduced blood glucose control. Specifically, the BMI percentiles of 5 of 8 participants were greater than the 85^{th} percentile, classifying them as obese or overweight. Moreover, impaired blood glucose is an associated symptom of metabolic disorders influenced by weight, such as metabolic syndrome and

type II diabetes mellitus. In these disease states, impaired blood glucose control may be evidenced by or prefaced by hyperinsulinemia and glucose intolerance (19, 20). This finding suggests blood glucose control in individuals with SMA type II may benefit from maintaining appropriate weight status.

Likewise, overfatness was prevalent among our participants, with the exception of 1 cachectic participant. As previously discussed, overfatness is an abnormal ratio of fat mass to fat free mass and is commonly seen in children with SMA when compared to healthy cohorts (14). When plotted according to NHANES smoothed body fat percentiles, 7 of 8 participants were classified above the 95%ile, the cut-off point for obesity, whereas when plotted according to CDC BMI-for-age charts, only 4 of 8 can be identified as obese and 1 participant as overweight. These results were also reflected in the findings of Hurst et al., where 6 of 6 participants were considered obese following DXA-determined body mass percentage and plotting on NHANES smoothed body fat percentiles, whereas only 3 participants identified as obese and 2 as overweight according to CDC BMI-for-age charts. Conclusively, this evidence further demonstrates BMI-for-age percentiles from CDC growth charts do not correlate with body composition for children with SMA. With this in mind, health professionals need to be aware that children with SMA type II are at higher risk for metabolic issues at a lower BMI due to differences in body composition compared to healthy peers. For example, children may have a normal BMI-for-age, yet their risk for metabolic-related issues may be increased. Use of body composition estimations, such as BIA or DXA, in addition to anthropometric measures, provides a more comprehensive assessment of metabolic health than anthropometrics alone in children with SMA type II.

Moreover, the incongruity of these measurement tools emphasizes the underlying idea that health professionals must stress the importance of weight control and tracking in children with SMA in order to proactively address issues and avoid complications of excess fat mass and obesity. These risks may be mitigated if children with SMA are able to work closely with a dietitian in order to address decreasing caloric need with loss of fat-free mass, decreased motor abilities, and other associated nutritional changes.

As a prospective exploratory study, our experimental design was simple and efficient. The CGM gathered comparable valid blood glucose information from each participant. These data had a strong correlation to actual blood glucose levels via information provided by daily fingersticks.

The study increased homogeneity of ingredients used during the prescribed specialized diet day by providing all nonperishables. Although we were not able to provide all foods due to cost and travel arrangements, we provided written and verbal instructions on preparation and provided a detailed menu. By giving discrete instructions and providing specified foods, we enabled the participants to consume as close to an identical diet for the prescribed specialized diet day, permitting for the most comparable data.

The design of our study does not greatly impede the daily life of the participant, increasing validity and insight into the day-to-day life and dietary patterns of a person living with SMA type II. The study had little participant responsibility aside from recording daily food intake and blood glucose fingersticks. The CGM allowed for data collection with few participant obligations and increased the validity of data collected. All participants in this study also participate in an ongoing longitudinal SMA study

which requires participants to submit daily food diaries. Therefore, recording dietary intake may not be seen as an additional obligation of the study.

Despite a strong study design, there are limitations. Our study relied on participant compliance to complete and return detailed daily food diaries to compare against the data collected by the CGM. Upon analysis using the ESHA software, some food diaries did not provide sufficient information to appropriately enter foods consumed and had to be estimated for brand, type, cooking style, and/or amount. These estimations may have altered the observed relationships between the blood glucose levels and dietary intake.

Due to limitations of the battery life of the CGM, participants had to complete all aspects of the study within 10 days. Therefore, the timespan of our intervention was limited and unfortunately only feasible for a single day. Long-term dietary interventions could have a more pronounced effect on health status and metabolic measures. The short-term effects seen, particularly with fiber intake, have potential to be more dramatic if practiced for a longer period of time.

Additionally, the blood glucose values of the nonspecialized diet days were evaluated as an average taken over 3 days, reducing the amplitude of blood glucose highs and lows experienced. This analysis could have potentially diminished the effects of the prescribed specialized diet day when compared. Therefore, the change seen during the prescribed specialized diet day, although not investigated for significance due to the small *n*, could have proven to have a greater impact if compared individually day-by-day to the nonspecialized diet days rather than as an average of the 3. Evaluation day-by-day instead of averaged values of the 3 nonspecialized days can be seen in Figures 2 and 3.

Our data also relied on the function and reporting of the CGM. For 1 participant, the CGM was at times unable to draw the necessary sample of blood; therefore, there were periods of time where no blood glucose values were reported. These time periods were removed from analysis.

The prescribed specialized diet day was designed to meet macronutrient intake needs and 75% of dietary recommended intake (DRI) for micronutrients. Personal experience with the tastes of the participant population encouraged us to choose pulp-free orange juice, a product with high naturally occurring sugars without notable fiber content, in place of milk to fulfill calcium and vitamin D requirements. As suspected, a notable spike in blood glucose followed by a greater drop was seen in participants following consumption of the juice. This blood glucose spike and decline may have mitigated the blood glucose control of the prescribed specialized diet day.

Another consideration for data analysis was that participant E was on overnight enteral feeds that consisted of a high carbohydrate, very low fat formula. This regimen resulted in blood glucose fluctuations overnight that were not seen in the rest of the study population and therefore may have mitigated the hypothesized control of the prescribed specialized diet day in the case of this participant.

Additionally, digression from protocol by a participant reduced sample size for the evaluation of the effects of the prescribed specialized diet day. Specifically, participant H was not compliant with protocol by consuming the prescribed specialized diet day meals over 2 days instead of 1 and did not completely abide by the prescribed food list. The second day, when the prescribed specialized diet was more thoroughly followed, was counted as the prescribed specialized diet day for analysis. This participant

was removed from Figure 1 due to these deviations.

Availability of laboratory results of blood draws was another limitation of our data analysis. HgA1c values for 1 participant were lost by the lab during testing. The participant did not complete this lab test outside of their appointment as recommended; therefore, only 7 data points were available for analysis concerning HgA1c.

Evaluation of insulin resistance using HOMA-IR scores can also be somewhat variable. Considering that scores are based on fasting insulin and fasting glucose levels, marked changes in these values can occur in a relatively short period of time, effecting HOMA-IR evaluation. For instance, in the study by Hurst et al., participants were tested twice for insulin resistance via HOMA-IR scores within 8 weeks. During the first visit, 4 of 6 participants were identified as insulin resistant, whereas at the second visit, 5 of 6 participants met the criteria. Additionally, HOMA-IR scores can be dependent on the judgment of the clinician as to whether or not the child is pre-pubertal. Dependent on this maturity classification, cut off values for HOMA-IR classification vary.

Additionally, our investigation focused on a small study population of 8 participants. The small population improved interpersonal care given to improve adherence to procedures and homogeneity. However, the limited number of participants decreased the strength of our analysis because the SMA type II population is very heterogeneous in terms of weight, mobility, respiratory function, and overall health status. Therefore, the small number of participants may not correlate with the breadth of the SMA type II population and limits the generalizability of the study.

Although dietary changes may improve blood glucose control, SMA disease progression may impact glycemic control to a greater extent than nutritional intervention.

However, the difference contributed by diet, as exemplified by this intervention, may provide a basis for further studies and clinical treatment of metabolic issues in individuals with SMA.

CONCLUSION

The outcomes of this study have the potential to elucidate associations between glycemic control, body composition, and SMA type II. The promising effects of improved glycemic control through increased fiber intake may be of interest for further studies and development of dietary interventions for SMA therapy. Likewise, outcomes of dietary intervention may have impact on participants with varying metabolic function and potentially be more significant if conducted for a longer period of time. Additionally, overfatness was common among participants and may correlate with impaired blood glucose control as BMI percentile increases. Lastly, body composition findings emphasize the inadequacies of using the CDC BMI-for-age growth charts to define overweight or obese status in children with SMA type II as well as the differences in body composition of children with SMA in comparison to healthy peers. Further investigation of extended dietary intervention influence and long-term metabolic changes in individuals with SMA type II is needed to determine relevance and effectiveness. Overall, the outcomes of this study elicit further investigation and awareness of nutritional differences within the SMA community.

APPENDIX

FOOD RECORD

FOOD RECORD

Child's name: _____

Name of the caregiver(s) completing this form: _____

Relationship to the child: _____

Date: _____

Phone contact information if needed for clarification:
Cell:_____
Home:_____
Work:_____

My child's feeds are:
___All by mouth
___By mouth and by feeding tube
___Restricted to feeding tube only

Please return this form to:

Rebecca Hurst Davis
Clinical Dietitian
Department of Neurology
University of Utah School of Medicine:
30 N. 1900 E, Room 3R149
Salt Lake City, Utah 84132
FAX: 801-587-9346
EMAIL: rhurst@genetics.utah.edu

Please contact any of our clinical coordinators for questions regarding completion of this form:
801-585-9717

Nutritional Care Guidelines for children with SMA and other neuromuscular disorders are available on our web site: http://medicine.utah.edu/neurology/research/swoboda

DIRECTIONS FOR KEEPING THIS FOOD DIARY:

Please record everything your child eats or drinks over a four day period, including the specialized diet day. This record should include feeds and supplements and all water or other liquids given via mouth, nasogastric, nasojejunal or gastrostomy tube. At the top of the page make sure to note the date and check the box of which day of the food record you are recording (Day 1, Day 2, Day 3, Day 4).

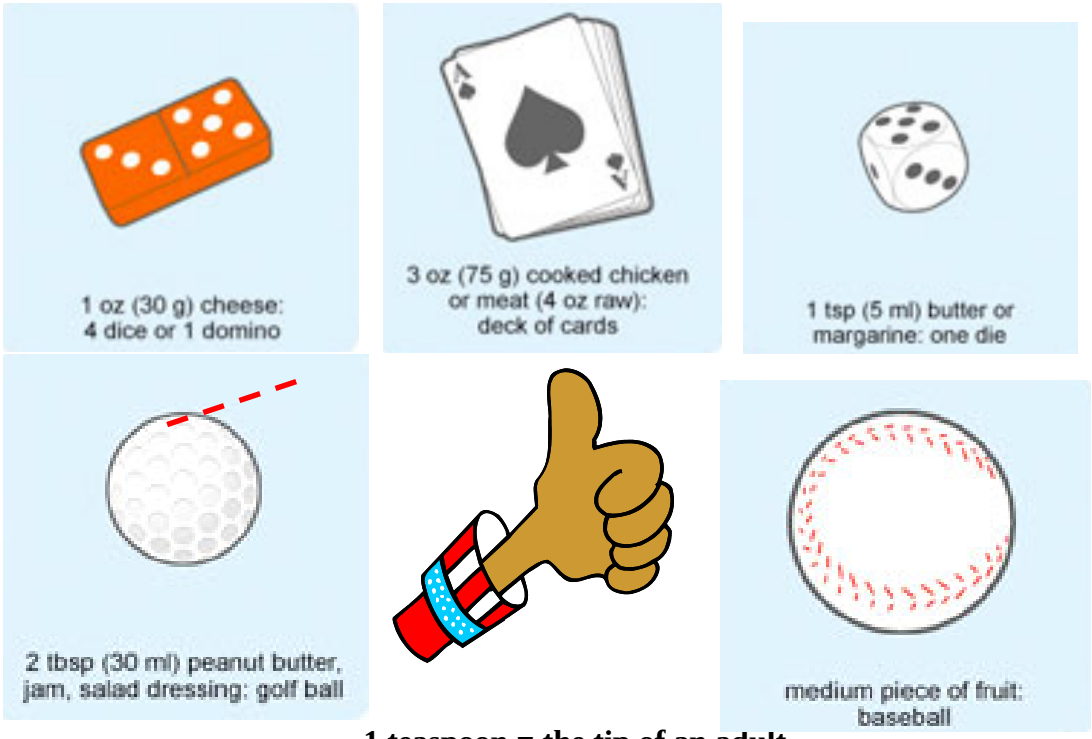
1. Record all food and drink whether eaten/given at home or away from home.
2. Be as specific as possible in recording the item, use a brand name if available.
3. Measure the item when possible or estimate the amount using the pictures provided with this document.
You can record in cubic centimeters (cc) or ounces (oz) with regard to liquids, and either ounces (oz) or grams (g) for food items.
4. Please mention how the food was prepared (fried, boiled, baked, microwaved, etc.) and record items used in the preparation, including oil, margarine or butter, for instance.
5. Don't forget to include all condiments, such as ketchup, gravy, sauce, butter or oil, and record amounts as accurately as possible. For instance, you might record 2 french fries with 1 teaspoon ketchup.
6. Please include one item per line where possible.
7. Please include all supplements (vitamins) given, with brand names and labels if possible.
8. You can use abbreviations for measurements:

Teaspoon – tsp ¼ cup – ¼ C Tablespoon – Tbsp Ounce – oz

9. Record blood glucose reading from glucometer finger stick in the "BG Value" column of the Food Diary. Finger sticks should be done 3-4 times daily. Suggested times for this are: before breakfast, before lunch, before dinner and at bedtime. Fingerstick glucose readings must be within 12 hours of each other.
10. In the exercise column please note any exercise completed. In the duration column, please note the length of time spent exercising.

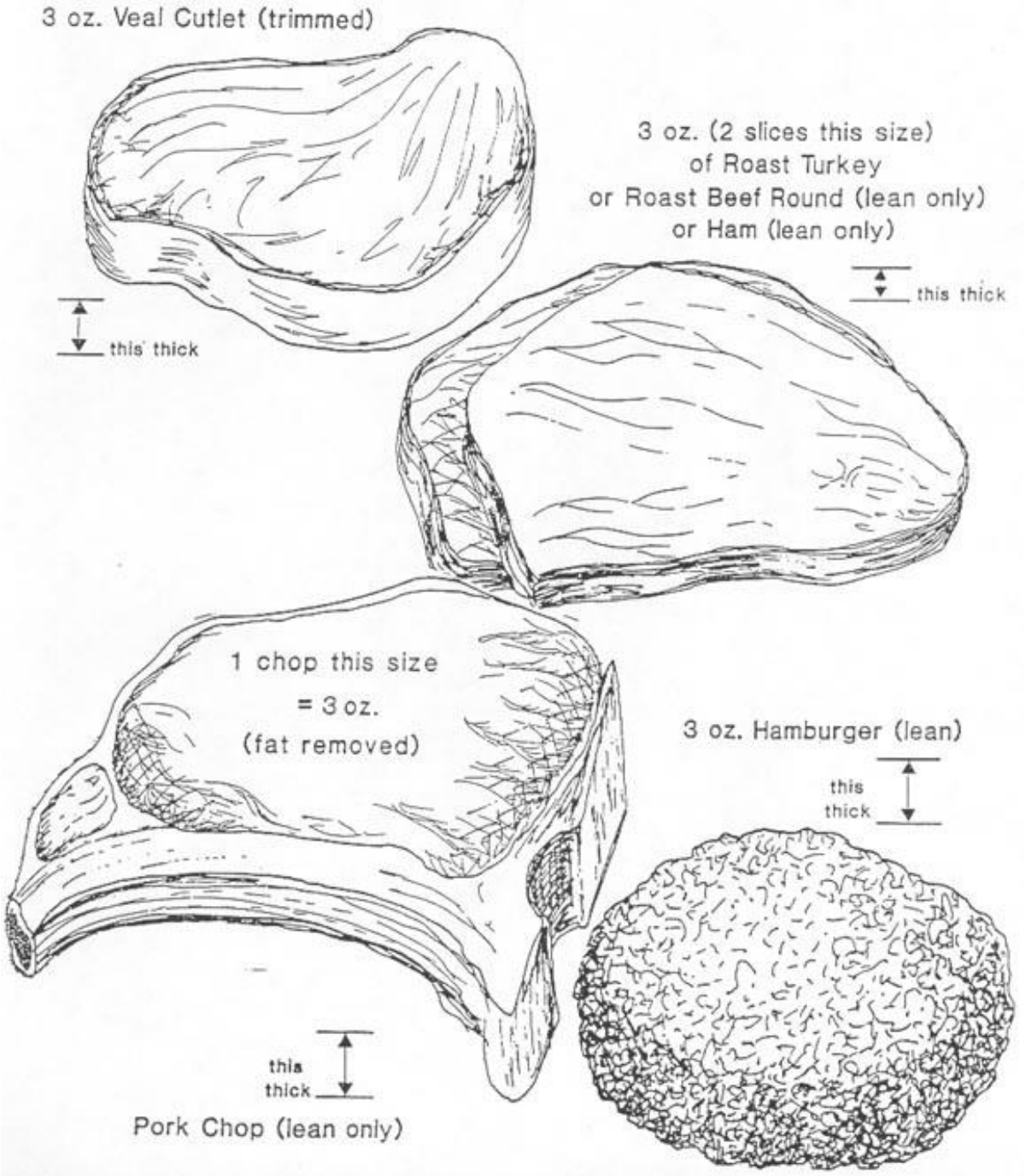
Time	BG Value	Food, Formula, or Fluid and Amount (include preparation details)	Exercise	Duration
7:00am	100			
7:30 am		½ c 2% milk		
		½ c Honey Nut Cheerios		
		1 medium banana		
8:00 am			Swimming	20 min

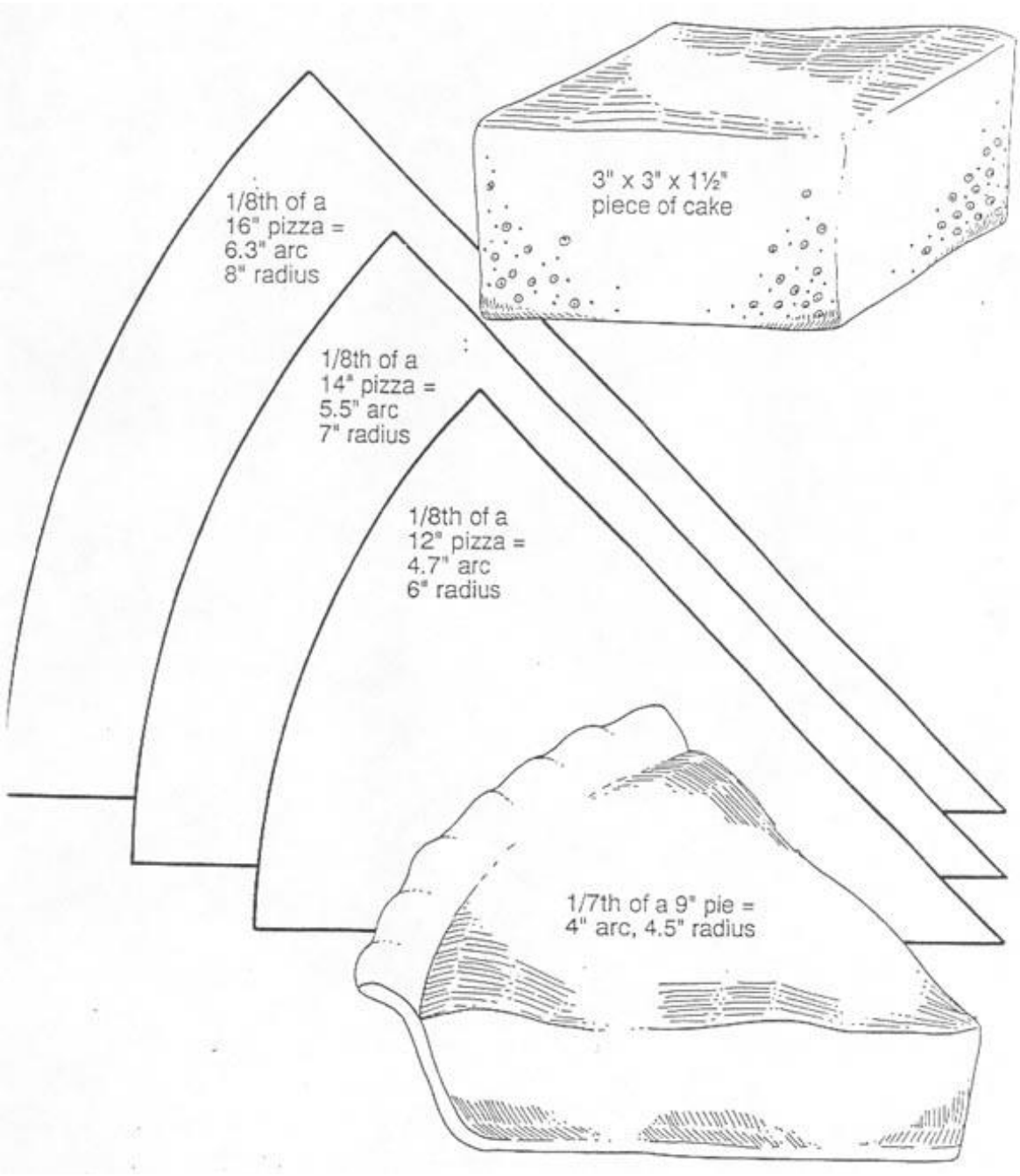
GUIDELINES FOR AMOUNTS OF WHOLE FOODS:



1 teaspoon = the tip of an adult thumb to the first joint

SERVING SIZES OF MEATS



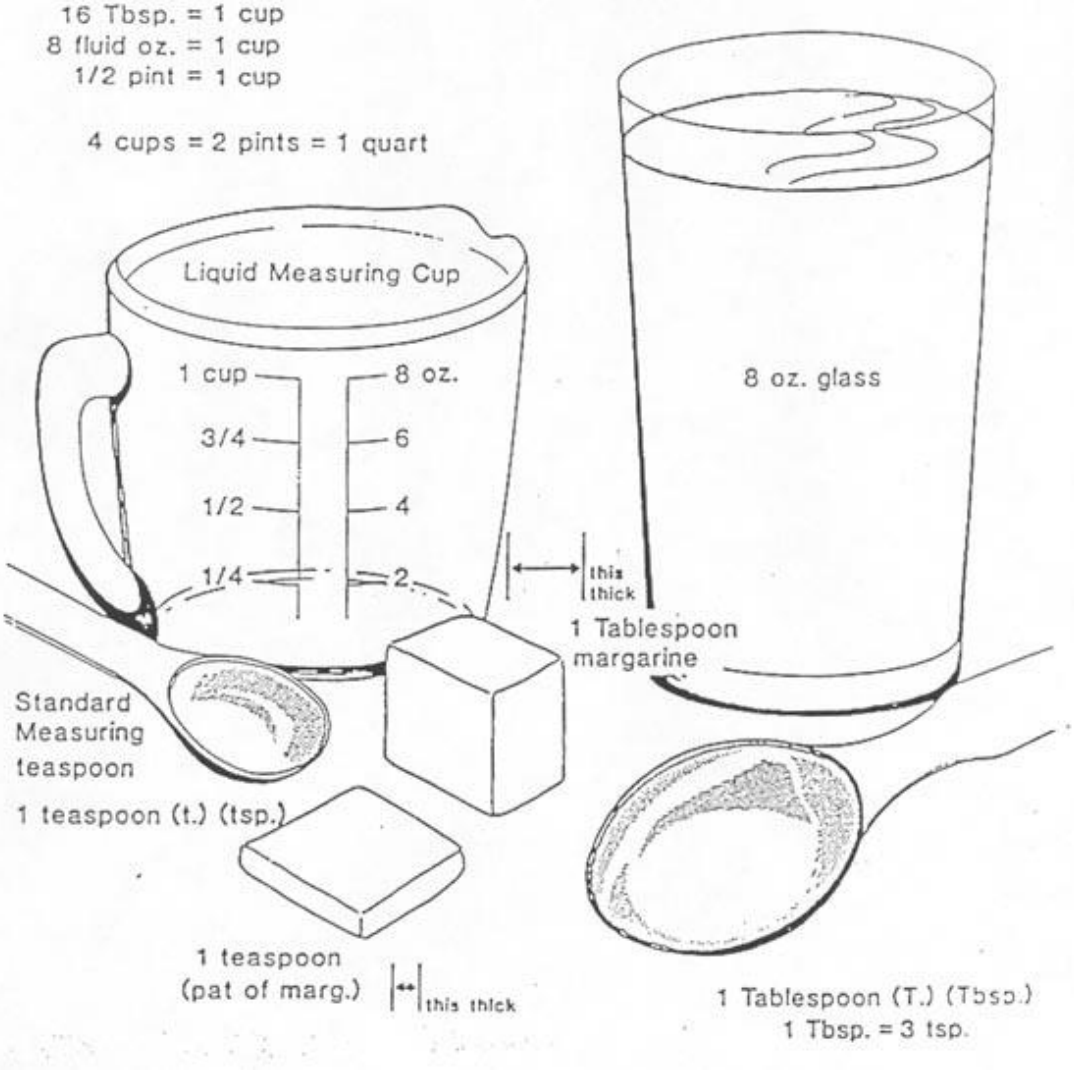

3" x 3" x 1½"
piece of cake
1/8th of a
16" pizza =
6.3" arc
8" radius
1/8th of a
14" pizza =
5.5" arc
7" radius
1/8th of a
12" pizza =
4.7" arc
6" radius
1/7th of a 9" pie =
4" arc, 4.5" radius

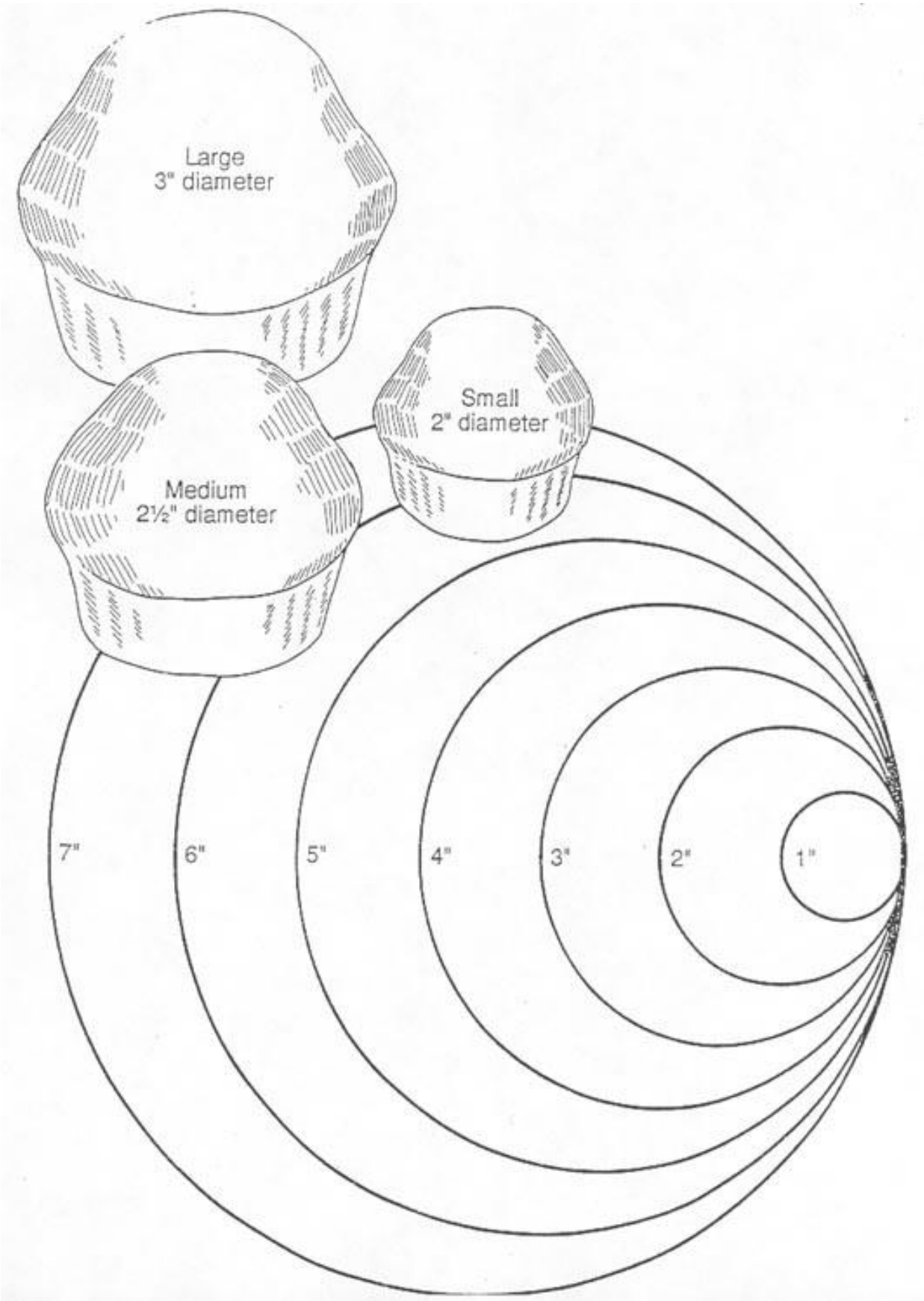
COMMON SERVING SIZES

4 Tbsp. = 1/4 cup
5 1/3 Tbsp. = 1/3 cup

16 Tbsp. = 1 cup
8 fluid oz. = 1 cup
1/2 pint = 1 cup

4 cups = 2 pints = 1 quart




Large
3" diameter
Small
2" diameter
Medium
2½" diameter
7"
6"
5"
4"
3"
2"
1"

Date: __________________________ Study ID:_______________________

iPro Serial Number: _______________ Day 1 [ ] Day 2 [ ] Day 3 [ ] Day 4 [ ]

Time	BG Value	Food, Formula, or Fluid and Amount (include preparation details)	Exercise	Duration

Date: ________________________ Study ID:_____________________

iPro Serial Number: _____________ Day 1 [ ] Day 2 [ ] Day 3 [ ] Day 4 [ ]

Time	BG Value	Food, Formula, or Fluid and Amount (include preparation details)	Exercise	Duration

Date: ________________________ Study ID:_____________________

iPro Serial Number: _____________ Day 1 [ ] Day 2 [ ] Day 3 [ ] Day 4 [ ]

Time	BG Value	Food, Formula, or Fluid and Amount (include preparation details)	Exercise	Duration

Date: __________________________ Study ID:_______________________

iPro Serial Number: _______________ Day 1 [ ] Day 2 [ ] Day 3 [ ] Day 4 [ ]

Time	BG Value	Food, Formula, or Fluid and Amount (include preparation details)	Exercise	Duration

Date: ________________________ Study ID:_____________________

iPro Serial Number: _____________ Day 1 [ ] Day 2 [ ] Day 3 [ ] Day 4 [ ]

Time	BG Value	Food, Formula, or Fluid and Amount (include preparation details)	Exercise	Duration

Date: ________________________ Study ID:_____________________

iPro Serial Number: _____________ Day 1 [ ] Day 2 [ ] Day 3 [ ] Day 4 [ ]

Time	BG Value	Food, Formula, or Fluid and Amount (include preparation details)	Exercise	Duration

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